

WHAT IS CLAIMED IS:

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C38T
1. A method of producing an immunogenic construct, comprising the steps of:

(a) activating at least one first carbohydrate-containing moiety with an organic cyanylating reagent to form an activated carbohydrate; and

(b) coupling said activated carbohydrate directly or indirectly to a second moiety to form an immunogenic construct capable of stimulating an immune response.

2. A method according to claim 1, wherein said organic cyanylating reagent is 1-cyano-4-(dimethylamino)-pyridinium tetrafluoroborate, N-cyanotriethyl-ammonium tetrafluoroborate, or p-nitrophenylcyanate.

2/3. A method according to claim 1, wherein said organic cyanylating reagent is 1-cyano-4-(dimethylamino)-pyridinium tetrafluoroborate.

Sub C4
4. A method according to claim 3, wherein said first and second moieties are soluble in water.

PD2 4/5. A method according to claim 3, wherein said activating step (a) is carried out at a pH of from 8 to 10, and said coupling step (b) is carried out at a pH of from 7 to 9.

5/6. A method according to claim 3, wherein said activating step (a) is carried out in the presence of triethyl amine.

6/7. A method according to claim 1, wherein the coupling in step (b) is done indirectly by covalently joining the ^{polysaccharide} first moiety to a bifunctional or heterofunctional spacer reagent, and covalently joining the ^{protein} second moiety to the spacer reagent.

C 7. ^{according to 6} A method ^{of claim 7}, wherein said spacer reagent is selected from the group consisting of ethylene diamine, 1,6-hexane diamine, adipic dihydrazide, cystamine, glycine, and lysine.

~~9. A method according to claim 1, wherein the first moiety is a polysaccharide and the second moiety is a protein~~

C 8. 10. A method according to claim ¹9, wherein the polysaccharide is selected from the group consisting of dextran, *Pneumococcal polysaccharide*, *Haemophilus influenzae* polysaccharide, Group A streptococcus polysaccharide, Group B streptococcus polysaccharide, and *N. meningitidis* polysaccharide.

C 9. 11. A method according to claim 1, wherein the ^{polysaccharide} ~~first moiety~~ is a water-soluble viral or bacterial polysaccharide.

C 10. 12. A method according to claim 1, wherein the ^{protein} ~~second moiety~~ is a water-soluble protein.

C 11. 13. A method according to claim 1, wherein the ^{protein} ~~second moiety~~ is selected from the group consisting of bovine serum albumin, pertussis toxoid, tetanus toxoid, malaria-derived peptide, an antibody, a toxoid, and a lipoprotein.

12. 14. A method according to claim 1, wherein the immunogenic construct is a conjugate selected from the group consisting of PT-Pn, PT-PRP, TT-Pn, antibody-dextran, and peptide-TT-Pn.

15. A method for producing an immune response, comprising:

(a) preparing an immunogenic construct by steps including (i) activating at least one first carbohydrate-containing moiety with an organic cyanylating reagent, and (ii) covalently joining said activated carbohydrate to a second moiety; and

(b) administering the immunogenic construct to a patient.

14 ~~16~~. A method according to claim ~~15~~¹³, wherein said organic cyanylating reagent is 1-cyano-4-(dimethylamino)-pyridinium tetrafluoroborate.

~~17~~. A ~~method~~ according to claim 16, wherein said activating step ~~(a)~~ is carried out in the presence of triethyl amine.

~~18~~. A method according to claim 16, wherein the first moiety is a polysaccharide and the second moiety is a water-soluble protein.

~~17~~¹⁴ ~~19~~. A method according to claim ~~18~~¹⁴, wherein the polysaccharide is selected from the group consisting of dextran, *Pneumococcal* polysaccharide, *Haemophilus influenzae* polysaccharide, Group A streptococcus polysaccharide, Group B streptococcus polysaccharide, and *N. meningitidis* polysaccharide.

~~18~~¹⁴ ~~20~~. A method according to claim ~~19~~¹⁴, wherein the ~~second~~^{protein} moiety is selected from the group consisting of bovine serum albumin, pertussis toxoid, tetanus toxoid, malaria-derived peptide, an antibody, a toxoid, and a lipoprotein.

19²¹. A method according to claim 16¹⁴, wherein the immunogenic construct is a conjugate selected from the group consisting of PT-Pn, PT-PRP, TT-Pn, antibody-dextran, and peptide-TT-Pn.